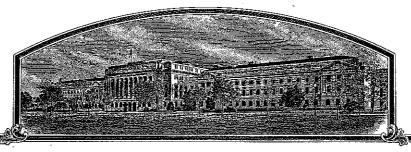
200000249

No.



THIE UNITED SHATES OF AMERICA

TO ALL TO WHOM THESE PRESENTS SHALL COME:

Enza Zaden Beheer B. A.

ALCCRIS, THERE HAS BEEN PRESENTED TO THE

Secretary of Agriculture

AN APPLICATION REQUESTING A CERTIFICATE OF PROTECTION FOR AN ALLEGED DISTINCT VARIETY OF SEXUALLY REPRODUCED, OR TUBER PROPAGATED PLANT, THE NAME AND DESCRIPTION OF WHICH ARE CONTAINED IN THE APPLICATION AND EXHIBITS, A COPY OF WHICH IS HEREUNTO ANNEXED AND MADE A PART HEREOF, AND THE VARIOUS REQUIREMENTS OF LAW IN SUCH CASES MADE AND PROVIDED HAVE BEEN COMPLIED WITH, AND THE TITLE THERETO IS, FROM THE RECORDS OF THE PLANT VARIETY PROTECTION OFFICE, IN THE APPLICANT(S) INDICATED IN THE SAID COPY, AND WHEREAS, UPON DUE EXAMINATION MADE, THE SAID APPLICANT(S) IS (ARE) ADJUDGED TO BE ENTITLED TO A CERTIFICATE OF PLANT VARIETY PROTECTION UNDER THE LAW.

NOW. THEREFORE, THIS CERTIFICATE OF PLANT VARIETY PROTECTION IS TO GRANT UNTO THE SAID APPLICANT(S) AND THE SUCCESSORS, HEIRS OR ASSIGNS OF THE SAID APPLICANT(S) FOR THE TERM OF TWENTY YEARS FROM THE DATE OF THIS GRANT, SUBJECT TO THE PAYMENT OF THE REQUIRED FEES AND PERIODIC REPLEMISHMENT OF VIABLE BASIC SEED OF THE VARIETY IN A PUBLIC REPOSITORY AS PROVIDED BY LAW, THE YIGHT TO EXCLUDE OTHERS FROM SELLING THE VARIETY, OR OFFERING IT FOR SALE, OR REPRODUCING IT, OR CORTING IT, OR CONDITIONING IT FOR PROPAGATION, OR STOCKING IT FOR ANY OF THE YE PURPOSE, OR CONDITIONING IT FOR PROPAGATION, OR STOCKING IT FOR ANY OF THE ABOVE SE, OR USING IT IN PRODUCING A HYBRID OR DIFFERENT VARIETY THEREFROM, TO THE EXTENT DBY THE PLANT VARIETY PROTECTION ACT. (84 STAT. 1542, AS AMENDED, 7 U.S.C. 2321 ET SEQ.)

LETTUCE

'Outback'

In Testimony Thereof, I have hereunto set my hand and caused the seal of the Hant Insisty Trotection Office to be affixed at the City of Washington, D.C. this twelfth day of December, in the year two thousand and five.

Atlast:

Dem Jel Commissioner

Commissioner
Plant Variety Protection Office
Agricultura! Marketing Service

MKGoboon Socrotary of Agriculturo

| REPRODUCE LOCALLY, include form number and de | ate on all reprodu | actions | | | | Form Approved - OMB No. 0581-0055 | | | |
|--|--|--|---|---|--------------------|--|--|--|--|
| U.S. DEPARTMEN AGRICULTURAL & | | | The following statements are made in accordance with the Privacy Act of 1974 (5 U.S.C. the Paperwork Reduction Act (PRA) of 1995. Application is required in order to determine if a plant variety protection certificate is to be (7 U.S.C. 2421). Information is held confidential until certificate is issued (7 U.S.C. 2422). | | | | | | |
| SCIENCE AND TECHNOLOGY - PI | | | | | | plant variety protection certificate is to be Issued | | | |
| APPLICATION FOR PLANT VAI (Instructions and information col | | | (7 U.S.C. 2 | 421). Information is held co. | nfidential | until certificate is issued (7 U.S.C. 2426). | | | |
| 1. NAME OF OWNER | | | 2. TEMPOI EXPERI | RARY DESIGNATION OR MENTAL NAME | 1 | RIETY NAME | | | |
| Enza Zaden Beheer B.V. | | | | | " | JTBACK | | | |
| 4. ADDRESS (Street and No., or R.F.D. No., City, | State, and ZIP Co | de, and Country) | 5. TELEPH | ONE (include area code) | \vdash | FOR OFFICIAL USE ONLY | | | |
| Postbus 7, Haling 1e | | | 00312 | 28350100 | PVPO | NUMBER | | | |
| 1600 AA , 1602 DB ENKHU | JIZEN | | 6. FAX (inc | lude area code) | 20 | 00000249 | | | |
| The Netherlands | | | 00312 | 28315960 | | DATE | | | |
| IF THE OWNER NAMED IS NOT A "PERSON", ORGANIZATION (corporation, partnership, associated) | | 8. IF INCORPORATED, GIVE STATE OF INCORPORATION | 9. DATE O | FINCORPORATION | , | -/10/11/00 | | | |
| Corporation | | Noord Holland | \mathbf{d} | 1936 | 5 | 5/18/2000 | | | |
| 10. NAME AND ADDRESS OF OWNER REPRESE | NTATIVE(S) TO S | L SERVE IN THIS APPLICATION. (First | person listed wi | il receive all papers) | F E | FILING AND EXAMINATION FEES: | | | |
| Mrs. Manon Knol | | | | | E S | : 2450.00 | | | |
| Enza Zaden Beheer B.V. Postbus 7, Haling 1e | | | | | R | DATE 5/18/2000 CERTIFICATION FEE: | | | |
| 1600 AA, 1602 DB ENKHUIZEN | | | | | E E | S CORD | | | |
| The Netherlands | | • | | | V E | 1 | | | |
| | | | | | 0 | DATE 11/3/05 | | | |
| 11, TELEPHONE (Include area code) | 12. FAX (includ | | 13. E-l | MAIL DI@enzazaden.nl | | | | | |
| 0031228350218 14. CROP KIND (Common Name) | 16. FAMILY NA | 28315960 AME (Botenicel) | | | IN ANY T | RANSGENES? (OPTIONAL) | | | |
| Lettuce | Lactuca sati | • | | | | • | | | |
| 15. GENUS AND SPECIES NAME OF CROP | 17. IS THE VAR | RIETY A FIRST GENERATION HYBR | ID? IF | SO, PLEASE GIVE THE AS | SSIGNED | USDA-APHIS REFERENCE NUMBER FOR THE ATE THE GENETICALLY MODIFIED PLANT FOR | | | |
| Lactuca sativa L. | YES | ☑ NO | 1 | OMMERICALIZATION | | | | | |
| 19. CHECK APPROPRIATE BOX FOR EACH ATTA (Follow instructions on reverse) | CHMENT SUBMI | TTED | | | | EED OF THIS VARIETY BE SOLD AS A CLASS 33(a) of the Plant Variety Protection Act) | | | |
| a. Exhibit A. Origin and Breeding History | of the Variety | | YES (If "yes", answer items 21 and 22 below) V NO (If "no", go to item 23) 21. DOES THE OWNER SPECIFY THAT SEED OF THIS VARIETY BE LIMITED AS TO | | | | | | |
| b. Exhibit B. Statement of Distinctness | | | NUMBER OF CLASSES? | | | | | | |
| c. Exhibit C. Objective Description of Varia | • | | | | D FOU | NDATION REGISTERED CERTIFIED | | | |
| d. Exhibit D. Additional Description of the e. Exhibit E. Statement of the Basis of the | | nin | 22. DO | ES THE OWNER SPECIFY | THAT S | EED OF THIS VARIETY BE LIMITED AS TO | | | |
| f. Voucher Sample (2,500 viable untreater | | | | IMBER OF GENERATIONS YES NO | • r | | | | |
| verification that tissue culture will be de repository) | posited and maint | ained in an approved public | | YES, SPECIFY THE NUMB | ER 1,2,3, | etc. FOR EACH CLASS. | | | |
| g. Filing and Examination Fee (\$3,652), m. States" (Mail to the Plant Variety Protect | | easurer of the United | | FOUNDATION RE | GISTERE | D CERTIFIED | | | |
| | | | (If a | additional explanation is nec | essary, p | lease use the space indicated on the reverse.) | | | |
| 23. HAS THE VARIETY (INCLUDING ANY HARVES FROM THIS VARIETY BEEN SOLD, DISPOSED OTHER COUNTRIES? | | | 24. IS 1 | THE VARIETY OR ANY CO TELLECTUAL PROPERTY | MPONEN RIGHT (1 | IT OF THE VARIETY PROTECTED BY PLANT BREEDER'S RIGHT OR PATENT)? | | | |
| YES NO | | | | YES 🗹 NO | | • | | | |
| IF YES, YOU MUST PROVIDE THE DATE OF F FOR EACH COUNTRY AND THE CIRCUMSTA | | | | ES, PLEASE GIVE COUNT ERENCE NUMBER. (Plea | | E OF FILING OR ISSUANCE AND ASSIGNED bace indicated on reverse.) | | | |
| 25. The owners declare that a viable sample of basic a tuber propagated variety a tissue culture will be | c seed of the varie e deposited in a p | ely has been furnished with application ublic repository and maintained for the | and will be repleted and will be repleted and will be repleted and all all and all all and all and all all all all all all all all all al | lenished upon request in ac certificate. | cordance | with such regulations as may be applicable, or for | | | |
| The undersigned owner(s) is(are) the owner of the entitled to protection under the provisions of Sec | nis sexually reprod tion 42 of the Plan | luced or tuber propagated plant variet it Variety Protection Act. | ariety, and believe(s) that the variety is new, distinct, uniform, and stable as required in Section 42, and is | | | | | | |
| Owner(s) is (ere) informed that false representat | | | enalties. | | | | | | |
| SIGNATURE OF OWNER LNZA | | | SIGNATURE OF OWNER | | | | | | |
| (tour | Postbus O AA En | zki i | | | | | | | |
| T | he Nether | | NAME (Please print or type) | | | | | | |
| R.J.P. Peerenboom | DATE | | CAPACITY OR TI | TLE | DATE | | | | |
| commercial director | | | commercial | | | 0/2005 | | | |
| Commercial director | 1 30/ | | Commeterat | director | | | | | |

U.S. DEPARTMENT OF AGRICULTURE AGRICULTURAL MARKETING SERVICE SCIENCE AND TECHNOLOGY - PLANT VARIETY PROTECTION OFFICE

The following state-nents are made in accordance with the Privacy Act of 1974 (5 U.S.C. 552a) and the Paperwork Reduction Act (PRA) of 1995.

APPLICATION FOR PLANT VARIETY PROTECTION CERTIFICATE

Application is required in order to determine if a plant variety protection certificate is to be issued (7 U.S.C. 2421). Information is held confidential until certificate is issued (7 U.S.C. 2426).

| (Instructions and information collection burden st | atement on rever | rse) | | | | |
|---|---|---|--|--|----------------|---|
| Name of owner Parker Vegetable Seeds E | nza Zaden B | Beheer B | .V. | 2. TEMPORARY DESIGNAT EXPERIMENTAL NAME | | 3. VARIETY NAME Outback |
| 4. ADDRESS (Street and No., or R.F.D. No., City, State, and ZIP Code | , and Country) | | | 5. TELEPHONE (include are | a code) | FOR OFFICIAL TISE ONLY |
| 13/19 Chifley Street | Gerrit Sasse | | DИ | 00.31.228.3513 | 18 | PVPO NUMBER |
| Smithfield 2164 AUSTRALIA | Enza Zaden Enkhuizen The Netherl | • | D.V. | 01007074000 | 3/105 | 20000002 FILING DATE |
| 7. IF THE OWNER NAMED IS NOT A "PERSON", GIVE FORM OF ORGANIZATION (corporation, partnership, association, etc.) Corporation | TATE | CORPORATEI TE OF INCORP V SOUTH | ORATION | 9. DATE OF INCORPORATION 9. DATE OF INCORPORAT | i | may 18,20 |
| 10. NAME AND ADDRESS OF OWNER REPRESENTATIVE(S) TO SEE | RVE IN THIS APPLICA | TION. (First pe | erson listed will rec | reive all papers) | | FILING AND EXAMINATION |
| Marcel Bruins Seminis Vegetable Seeds, Inc. Nude 54D 6702 DN Wageningen The Netherlands | Mr. D. S 13/19 Ch Smithfie Australia | nifley St ld 2164 | | 2AD 1/25/05 | | FEES: \$ 2450. — BATE MAY 18, 20 CERTIFICATION FEE: DATE DATE |
| 11. TELEPHONE (Include area code) 12. FAX (Include area c | code) | 13. E_MAR | | | 14. CROP | KIND (Common Name) |
| 612 97 25 12 00 fax 612 97 2 | 25 10 66 | mbri | Jins@ s | vseeds.nl | Let | tuce |
| 15 GENUS AND SPECIES NAME OF CROP | · | | Y NAME (Bolanica | | | VARIETY A FIRST GENERATION |
| Lactuca sativa | : | 1 | raceae | | HYBRI | 0?] YES \[\](NO |
| 18. CHECK APPROPRIATE BOX FOR EACH ATTACHMENT SUBMITTS reverse) a. Exhibit A. Origin and Breeding History of the Variety b. Exhibit B. Statement of Distinctness c. Exhibit C. Objective Description of Variety d. Exhibit D. Additional Description of the Variety (Optional) | ED (Follow instructions | 5 001 | CERTIFIED S | EED? See Section 83(a) of ES (If "yes", answer items 20 and 21 below) MNER SPECIFY THAT SEED (| the Plant Var | IETY BE SOLD AS A CLASS OF iety Protection Act) [K NO (II "no," go to item 22) IETY BE LIMITED AS TO NUMBER |
| e. 🗸 Exhibit E. Stalement of the Basis of the Owner's Ownersh | • | | ☐ YE | :S | E | |
| Voucher Sample (2,500 viable untreated seeds or, for tube verification that tissue culture will be depositied and maintain repository) 9. V Filing and Examination Fee (\$2,450), made payable to "Tri States" (Mail to the Plant Variety Protection Office) | | i, ublic 2 | _ | TEM 20, WHICH CLASSES OF DUNDATION | | N BEYOND BREEDER SEED? CERTIFIED |
| 22. HAS THE VARIETY (INCLUDING ANY HARVESTED MATERIAL) OF FROM THIS VARIETY BEEN SOLD, DISPOSED OF, TRANSFERRE | A HYBRID PRODUC | ED 2 | 3. IS THE VARIE | TY OR ANY COMPONENT OF IGHT (PLANT BREEDER'S RIC | THE VARIET | Y PROTECTED BY INTELLECTUAL |
| OTHER COUNTRIES? XX YES 5/21/99 USA no if YES, YOU MUST PROVIDE THE DATE OF FIRST SALE, DISPO- FOR EACH COUNTRY AND THE CIRCUMSTANCES. (Please use | | | ☐ Y | ES E GIVE COUNTRY, DATE OF NUMBER. (Please use space in | FILING OR IS | NO SSIGNED |
| 24. The owners declare that a viable sample of basic seed of the variety for a tuber propagated variety a tissue culture will be deposited in a part of the undersigned owner(s) is(are) the owner of this sexually reproduce and is entitled to protection under the provisions of Section 42 of the I Owner(s) is(are) informed that talse representation herein can jeopan | oublic repository and m ed or tuber propagated Plant Variety Protection | mintained for the d plant veriety, n Act. | ne duration of the c and believe(s) tha | certificate. | - | |
| SIGNATURE OF OWNER | | | SIGNATURE OF O | WNER | | |
| NAME (Please print or type) Yates Vegetable Seeds / DANIEL | TRIMB | | IAME (Please prin | f or type) | | |
| | _{оате} 9-Мау-00 | | CAPACITY OR TIT | LE . | | DATE |
| AT-470 (5-98) designed by the Plant Variety Protection Office with WordPe | rfect 6.0a. Replaces | STD-470 (03-9 | 6) which is obsole | e. (See reverse for inst | ructions and i | nformation collection burden statement |

200000249

INSTRUCTIONS

GENERAL: To be effectively filed with the Plant Variety Protection Office (PVPO), ALL of the following items must be received in the PVPO: (1) Completed application form signed by the owner; (2) completed exhibits A, B, C, E; (3) for a seed reproduced variety at least 2,500 viable untreated seeds, for a hybrid variety at least 2,500 untreated seeds of each line necessary to reproduce the variety, or for tuber reproduced varieties verification that a viable (in the sense that it will reproduce an entire plant) tissue culture will be deposited and maintained in an approved public repository; (4) check drawn on a U.S. bank for \$3,652 (\$432 filing fee and \$3,220 examination fee), payable to "Treasurer of the United States" (See Section 97.6 of the Regulations and Rules of Practice.) Partial applications will be held in the PVPO for not more than 90 days, then returned to the applicant as unfiled. Mail application and other requirements to Plant Variety Protection Office, AMS, USDA, Room 401, NAL Building, 10301 Baltimore Avenue, Beltsville, MD 20705-2351. Retain one copy for your files. All items on the face of the application are self explanatory unless noted below. Corrections on the application form and exhibits must be initiated and dated. DO NOT use masking materials to make corrections. If a certificate is allowed, you will be requested to send a check payable to "Treasurer of the United States" in the amount of \$432 for issuance of the certificates will be issued to owner, not licensee or agent.

Plant Variety Protection Office Telephone: (301) 504-5518 FAX: (301) 504-5291

Homepage: http://www.ams.usda.gov/science/pvpo/pvpindex.htm

To avoid conflict with other variety names in use, the applicant must check the appropriate recognized authority and provide evidence that name has been cleared by the appropriate recognized authority before the Certificate of Protection is issued. For example, for agricultural and vegetable crops, contact: Seed Branch, AMS, USDA, 10301 Baltimore Avenue, Suite 401 NAL Building, Beltsville, MD 20705. Telephone: (301) 504-5682 http://www.ams.usda.gov/lsg/seed.htm.

ITEM

19a. Give:

- (1) the genealogy, including public and commercial varieties, lines, or clones used, and the breeding method;
- (2) the details of subsequent stages of selection and multiplication;
- (3) evidence of uniformity and stability; and
- (4) the type and frequency of variants during reproduction and multiplication and state how these variants may be identified
- 19b. Give a summary of the variety's distinctness. Clearly state how this application variety may be distinguished from all other varieties in the same crop. If the new variety is most similar to one variety or a group of related varieties:
 - (1) identify these varieties and state all differences objectively;
 - (2) attach statistical data for characters expressed numerically and demonstrate that these are clear differences; and
 - (3) submit, if helpful, seed and plant specimens or photographs (prints) of seed and plant comparisons which clearly indicate distinctness.
- 19c. Exhibit C forms are available from the PVPO Office for most crops; specify crop kind. Fill in Exhibit C (Objective Description of Variety) form as completely as possible to describe your variety.
- 19d. Optional additional characteristics and/or photographs. Describe any additional characteristics that cannot be accurately conveyed in Exhibit C. Use comparative varieties as is necessary to reveal more accurately the characteristics that are difficult to describe, such as plant habit, plant color, disease resistance, etc.
- 19e. Section 52(5) of the Act requires applicants to furnish a statement of the basis of the applicant's ownership. An Exhibit E form is available from the PVPO.
- 20. If "Yes" is specified (seed of this variety be sold by variety name only, as a class of certified seed), the applicant MAY NOT reverse this affirmative decision after the variety has been sold and so labeled, the decision published, or the certificate issued. However, if "No" has been specified, the applicant may change the choice. (See Regulations and Rules of Practice, Section 97.103).
- 23. See Sections 41, 42, and 43 of the Act and Section 97.5 of the regulations for eligibility requirements.
- 24. See Section 55 of the Act for instructions on claiming the benefit of an earlier filing date.
- 22. CONTINUED FROM FRONT (Please provide a statement as to the limitation and sequence of generations that may be certified.)
- 23. CONTINUED FROM FRONT (Please provide the date of first sale, disposition, transfer, or use for each country and the circumstances, if the variety (including any harvested material) or a hybrid produced from this variety has been sold, disposed of, transferred, or used in the U.S. or other countries.)
 5/21/1999 USA
- 24. CONTINUED FROM FRONT (Please give the country, date of filing or issuance, and assigned reference number, if the variety or any component of the variety is protected by intellectual property right (Plant Breeder's Right or Patent).)

NOTES: It is the responsibility of the applicant/owner to keep the PVPO informed of any changes of address or change of ownership or assignment or owner's representative during the life of the application/certificate. The fees for filing a change of address; owner's representative; ownership or assignment; or any modification of owner's name is specified in Section 97.175 of the regulations. (See Section 101 of the Act, and Sections 97.130, 97.131, 97.175(h) of the Regulations and Rules of Practice.)

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 0581-0055. The time required to complete this information collection is estimated to average 1.4 hours per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information.

The U.S. Department of Agriculture (USDA) prohibits discrimination in all its programs and activities on the basis of race, color, national origin, gender, religion, age, disability, sexual orientation, marital or family status, political beliefs, parental status, or protected genetic information. (Not all prohibited bases apply to all programs.) Persons with disabilities who require alternative means for communication of program information (Braille, large print, audictape, etc.) should contact USDA's TARGET Center at 202-720-2600 (voice and TDD).

To file a complaint of discrimination, write USDA, Director, Office of Civil Rights, Room 326-W, Whitten Building, 14th and Independence Avenue, SW, Washington, DC 20250-9410 or call 202-720-8964 (voice and TDD). USDA is an equal opportunity provider and employer.

Origin and Breeding History of Outback

In October 1994 the cross (F₁) between Green Towers (female) and the breeding line Lobjoits x LE 075 (male and source of resistance to corky root) was made. The F₁ was selfed and its progeny (F₂) screened for resistance to corky root. Resistant plants were backcrossed to Green Towers. Prior to each subsequent backcross, selfing of each backcrossed plant occurred and resistant progeny identified as listed above. The total number of backcrosses was three. The final backcross was then selfed and one resistant progeny selected at random. This plant (#4) was selfed and carried on for field evaluation.

This breeding was carried out at the Yates Research Station, N.S.W., Australia by Mr. D. S. Trimboli.

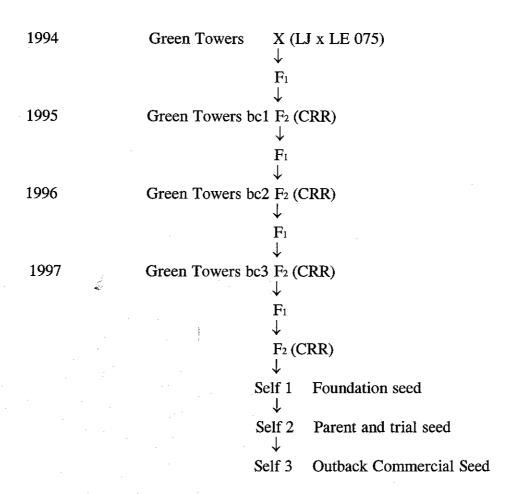


Exhibit A

Selection and Multiplication

A single F2 plant (#4) originating from selfing the F1 of backcross three was found to be resistant to corky root. Its progeny, Self 1, was multiplied in the field & Self 2 used for trials in California in 1998.

No vigorous selection was required in Outback because of the number of backcrosses used in the breeding. This was evident in Self 2 generation, which was novel and uniform and therefore used as parent seed. This produced commercial seed in the 1998/9 seed production season in Australia. The commercial seed is Self 3.

Uniformity/Distinctness /Stability

The S1, S2 and S3 progenies of Outback have shown uniform phenotypic characteristics for each generation. In addition these three generations were resistant to the corky root bacterium (*Rhizomaonas suberfiaciens* – N.S.W. Agriculture Herbarium # DAR69823) – in greenhouse tests at Narromine, Australia in April 1999.

Similarly the three generations have shown to be resistant to corky root in field trials in California.

The resistance to corky root in each generation indicates that Outback is distinct compared to the comparator variety, Green Towers, which is susceptible to corky root. Outback therefore, is uniform and stable for phenotype & disease resistance over three generations.

Variants

No variants as mutations have been observed in Outback. However, natural outcrosses can randomly occur but at a frequency of < 0.1%.

Exhibit B

Novelty Statement of Outback

Outback is a tall, robust, medium-green romaine lettuce belonging to the Parris Island Cos class. It is most similar to the commercial variety Green Towers.

When trialled and assessed for phenotypic characteristics, Outback produced a seed stalk more slowly than Green Towers at 55.4 and 53.7 days in 1999 and 64.3 and 62.2 days in 2000 respectively.

When trialled in Australia in 2000, bolting plants of Outback had a greater diameter than Green Towers at 36.7 and 34.2cm respectively. When trialled in the US, Outback had a lower core height than Green Towers at 71.6 and 80.0mm respectively.

The major difference between Outback and Green Towers is that Outback is resistant to corky root rot (*Rhizomonas suberifaciens*) and Green Towers is susceptible. This resistance contributes enormously to Outback's uniformity, earliness of maturity, height and head weight compared to Green Towers when both are grown in soil infested with the corky root bacterium. The differences between Outback and Green Towers are greater, proptionately to the severity of corky root in the field.

The resistance of Outback to corky root over two generations has been verified by DNA/molecular marker technology at Enza Zaden, Enkhuizen, Netherlands on 24th May, 2005. The marker is closely linked to the corky root resistance gene as described by:

Moreno-Vazquez, S. et al. (2003). SNP-based codominant markers for a recessive gene for conferring resistance to corky root rot (Rhizomonas suberifaciens) in lettuce (Lactuca sativa). Genome, <u>46</u>: 1059-1069.

Seed was used as the source of DNA and seedlots Outback- stockseed and Outback – commercial seed, represent generation one and two of Outback respectively. The number of seeds tested per seedlot was 36.

The results indicate that each Outback generation is homozygous resistant (RR) to corky root which clearly indicates repeatability between generations. The comparator, Green Towers, is homozygous recessive (susceptible – rr) for the corky root gene, indicating the distinctness of Outback compared to Green Towers. Gladiator, Augustus, King Henry

and E16.LE157 are resistant romaine varieties and the results confirm its homozygous resistance to corky root. Green Forest is also a susceptible romaine and the test confirmed this. This information is listed in Figure 2A and 2B. The results in Figure 2B are expressed in a microtitre plate format.

Therefore Outback is suited to lettuce growing areas especially where corky root is a problem. These areas include coastal California during summer and fall as well as northeastern states such as New Jersey.

Evaluation of Characteristics of Outback and Green Towers

Note: It was decided to conduct the Australian and American trials in soil not infested with corky root because plants grown in corky root-infested soil would perform poorly and inconsistently depending upon the severity of corky root in the soil. This applies essentially to the susceptible variety whose characteristics would be detrimentally affected by the disease. As levels of corky root vary in soil it would be difficult to reproduce trial results consistently when the severity of corky root in the soil cannot be quantified.

Owing to timing, both bolting trials had to be carried our in Australia.

The data are listed in Table 1.

Table 1: Evaluation of Characteristics of Outback and Green Towers in Australia and United States

| | Aus | stralia | United | d States |
|----------------------------------|-----------|--------------|----------|--------------|
| | Outback | Green Towers | Outback | Green Towers |
| Spread of Frame Leaves (cm) | | | | |
| Mean | 54.6 | 52.9 | 40.5 | 39.3 |
| Variance | 1.6 | 4.3 | 3.19 | 3.55 |
| t-value | -2.2090 | | -1.7394 | |
| Head diameter (cm) | | | | |
| Mean | 30.1 | 29.6 | 16.8 | 16.3 |
| Variance | 1.43 | 1.82 | 2.87 | 3.8 |
| t-value | -0.8763 | | -0.799 | |
| Head weight (g) | | | | |
| Mean | 667.5 | 590 | 956.6 | 1015 |
| Variance | 2090.2 | 8222.2 | 16916.66 | 5517.85 |
| t-value | -2.4133 | | 1.5083 | |
| Core height (mm) | | | | |
| Mean | 49 | 47.5 | 71.6 | 80 |
| Variance | 4.88 | 33.3 | 30.54 | 37.06 |
| t-value | -0.7666 | | * 3.9879 | |
| Maturity (days) | | | | |
| Mean | 52.1 | 52.5 | 94.2 | 93.5 |
| Variance | 0.62 | 2.36 | 1.17 | 1.12 |
| t-value | 1.0346 | | -1.7042 | |
| | 1999 | 1999 | 2000 | 2000 |
| No. days to seed stalk emergence | | | | |
| Mean | 55.4 | 53.7 | 64.3 | 62.2 |
| Variance | 2.19 | 2.3 | 1.22 | 3.62 |
| t-value | * -5.4448 | | *-4.6299 | |
| Height of mature seed stalk (cm) | | | | , |
| Mean | 85.9 | 84.9 | 120.7 | 120.7 |
| Variance | 21.75 | 16.59 | 17.41 | 8.85 |
| t-value | -1.119 | | -0.0101 | |
| Spread of bolter plant (cm) | | | | |
| Mean | 42.9 | 40.6 | 36.7 | 34.2 |
| Variance | 13.2 | 4.34 | 6.21 | 3.35 |
| t-value | -2.4551 | | *-3.161 | |

^{* =} significant at 0.01

Note: trial conditions must have varied between sites and seasons, causing the results not to be duplicated in either country.

RR resistant

RR resistant

FIGURE 2A

CORKY ROOT MARKER PLATE RESULTS

Outback stockseed - Generation 1

Outback commercial seed – Generation 2 RR resistant

Green Towers rr susceptible

Green Forest rr susceptible

E16.LE157 RR resistant

King Henry RR resistant

Gladiator RR resistant

24th May, 2005

Augustus



Overview of to be tested microtiterplate

Numbers of plates to be tested:

| | | | | : <u>·</u> | 1 | | | Ţ | | | Ī | | | | T | Т |
|--------------------------------|------------------------|------------------------|------------|------------|----|---|---|----|---|----|---|---|---|-----------------|---|---|
| System Sample | | | | | | | | | | | | | | | | |
| Harvest | 24-05-2005 | 24-05-2005 | 24-05-2005 | | | 1 | | | | | | | | | | |
| Hogram | CR toets voor Kees/Dan | CR toets voor Kees/Dan | FIGURE 2 B | | | | | | | | | | | | | |
| To be tested with the markers. | CR | 85 | 1 1 | | | | | | | | | | | | | |
| encount . | NL 05-402 | NL 05-403 | NL 05-404 | | - | | | | | | | / | | | | |
| :səɪvɪd | _ | 2 | 3 | 4 | S. | 9 | 7 | 90 | 6 | 10 | | | - | | | |

| ENZAZADEN | | 0 | | _ | _ | | - | | | | | | | | |
|------------------------------|----------------------------|-------------------------|--------|--|--------|----------|---------------|-----------|--------|--------|----------|---------|----------|---------------------------------------|----------------------------------|
| Date harvest: 24 EUS-2005 | | Date: 00-00-0000 | | | | | | | | | | | | | |
| × | | Õ | 8 | | ₩ ₩ | 2 | RR | RR | KZ X | RR. | ₩ | RR | RR | ₹ <u></u> | æ |
| vest: | 1 | | # | - 6 | -2 | 77 | 4 | 1.2 | φ | -7 | 89 | 6, | 9- | ÷ | -12 |
| Date har | DNA-check: yes/no | | Plant# | | | | | | | | | | | | |
| | heck: | | 2 | <u> </u> | 7 | m | 4 | w | 9 | 7 | ∞ | 9 | 2 | . = | 12 |
| | NA-c | ed by | | | | | | | | | | | | | |
| | | Results scored by: | | | | | | - | | | | | - | - | |
| | | Resul | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | |
| | | | _ | | | | | | | | | | | | |
| | ည | | 8 | -24 RR | -25 RR | -26 RR | -27 RR | -28 RR | -29 RR | -30 RR | -31 RR | -32 RR | -33 RR | -34 RR | <u> </u> |
| | CTAB/Promega/Kac | | Plant# | , | | 7 | \ \frac{1}{2} | 17 | ۲۰ | | | | 1 | \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ | Outback commercial RR seed |
| | B/Pro | | C | 1 | 7 | 6 | 4 | w | 9 | 7 | 20 | 6 | 10 | Ξ | 12 |
| | CTA | | | | | | | | | | | | | | |
| | | | | | | | | - | | | | | | | |
| | A-isolation: | | | | | | | | | | | | | ļ | |
| | isola | | | | | | | | | | | | | | |
| | DNA- | | | | | | | | | | | | | | |
| | - | | S. | | RR | RR | RR | * | RR. | RR | XX. | RR | 8 | ₩ | XX XX |
| nces: | 1.0 | | | | -13 | -14 | -15 | 91- | -17 | -18 | -19 | -50 | -21 | -22 | -23 RR |
| Resistances: | 412 | | Plant# | | | | | | | | | • | | | |
| | A1 - H12 | | В | 1 | 7 | m | 4 | ın. | 9 | 7 | 90 | • | 10 | 11 | 12 |
| A | 7 | | | | | • | | | | | | | • | 1 | |
|) } } | :: | | | | | | | | | | | | | | |
| 0.7 | sition | Pents | | | | | | | | | | | | | |
| 12 | If seeds, which positions: | R roels) (one Kees/D) m | | | | | | | | | | | | | |
| [E # | s, whi | 10001 | | | | | | | | | | | | | |
| 'LA'I | seeds | K toet | క | RR. | RR | RR | RR | RR | RR | RR | RR | RR | ~ | R | . ₂₄ |
| ER | ΙΕ | | | | 7 | -3 | 4- R | . 5 R | -6 R | -7 R | ∞- 22 | -9 R | -10 RR | -11 RR | -12 RR |
| MICROTITERPLATE # NIL 05-402 | :: | : : | Plant# | Outback stockseed | | | | | | | | - | , i | • | · |
| ICR | Material: | Program: | | · U) | | | | | | | | | | | |
| Z | ۶ الخ | 扎 | ¥ | *** | 7 | 6 | 4 | 4C | 9 | 7 | ေ | ٥ | 10 | 11 | 12 |

| | ļ · | 1 | | | | | | | | [| | |
|---------------|-------------|--------|--------|----------|--------|--------|--------|--------|--------|--------|--------|--------------|
| | | | | | | | | | | | | |
| | | | | | | | | | | | | |
| చ | | -14 п | -15 п | -16 п | -17 ш | -18 II | -19 | -20 rr | -21 п | -22 II | -23 п | -24 II |
| Plant# | | | | | | | - | | | | | |
| Ξ | 1 | 7 | 6 | 4 | ın | 9 | 7 | 90 | 6 | 10 | 11 | 17 |
| | | | | | | | | | | | | |
| | | | | | | | | | | | | á. |
| | | | | | | * | | | | | | |
| CR | Ħ | ㅂ | E | E | E | ㅂ | ㅂ | 궕 | 꿃 | 꾮 | Ħ | E |
| Plant# | -2 | £• | 4 | ý | 9- | 2- | ∞- | 6- | -10 | 17- | -12 | -13 |
| Ů | 1 | 7 | m | 4 | ın | 9 | 7 | ∞ | ٥ | 10 | 11 | 12 |
| | | | | | | | | | | | - / | |
| | | | | | | | | | | | | |
| | | | | | | | | | | | | |
| ₂₄ | | 24 | ~ | <u>ہ</u> | | æ | gy. | ~ | ~ | 22 | ı | L |
| Plant# CR | 建筑设置 | -25 RR | -26 RR | -27 RR | -28 RR | -29 RR | -30 RR | -31 RR | -32 RR | -33 RR | -34 Rr | Green Forest |
| В | 1 | 7 | ю | 4 | ıç. | 9 | 7 | ∞ | 6 | 10 | 11 | 12 |
| | | | | | | | | | | | | |
| | | | | | | | | | | | | |
| | - | | | | · | | | | | | | |
| రో | æ | RR | RR | RR | RR | RR | RR | RR | X | RR. | RR | æ |
| Plant# | -13 | -14 | -15 | -16 | 17 | -18 | -19 | -20 | -21 | -22 | -23 | -24 |
| | | | | | | | | | | | | |

| NEGNZVZNE | NL 05- | | | | | | | | | | | | | | | | | | | | | - | | | | | | | |
|-----------------------------|---------------------------|--|--------|--------------|-------|-------|-------|-------|-------|-------|-------|------|----------|------------|-------|---|---------|-------|------|-------|----------|------------|-------|--------|------|-------|----------|----------|--------|
| 11,787,773 | 7.53 | Date: 00-00-0000 | | | | | | | | | | | | | | | | | | | | , | | | | | | | |
| 4.4c | | Da | ž | | 뙶 | 뙲 | RR | RR | RR. | æ | R.R. | RR. | XX. | RR | RR | | CR | | RR | RR. | RR | RR. | RR | RR | RR. | RR | RR | 5 | 2 |
| Date harvest. 241.05-2005 | yes / no | | Plant# | | -26 | -27 | -28 | -29 | -30 | -31 | -32 | -33 | -34 | King Henry | -2 | | Piant# | | 4 | -5 | 9- | <i>L</i> - | 80 | 6- | -10 | -11 | -12 | -13 | -14 |
| | ieck: | | a | - | 7 | ю | 4 | 5 | 9 | 7 | ∞ | 6 | 10 | 11 | 12 | | Ħ | 1 | 7 | 6 | 4 | 5 | 9 | 7 | 8 | 6 | 10 | 11 | 12 |
| | DNA-check: yes/no | Results scored by: | | | | | | | | | | | | | | | | | | | | | | - | | | - | - | |
| | | | ₩ ₩ | ٠. | RR. | X. | RR | RR | #2 | RR | X3 | 22 | RR RR | R. | RR | : | S. | RR | RR | RR | RR | RR | RR | RR | RR | RR | RR | RR | RR |
| | a/Kac | 2000 1000 2000 | | -14 | -15 | -16 | -17 I | -18 | -19 | -20 | -21 E | -22 | -23 | -24 | -25 E | | | -26 F | -27 | -28 F | -29 | -30 | -31 F | -32 I | -33 | -34 I | Augustus | -2- | -3 E |
| | CTAB/Promega/Kac | | Plant# | | | | - | | | : | | | | | | | Plant# | | | | | | | | | | Aug | | |
| | LAB/F | | Ü | 1 | 2 | 3 | 4 | S | 9 | 7 | 8 | 6 | 10 | 11 | 12 | | G | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 6 | 10 | 11 | 12 |
| 3 | DNA-isolation: | | | | | | | | | | • | | - | | | | | | | | - | | | | | | | | |
| | | | GR | Secretario | -3 RR | -4 RR | -5 RR | -6 RR | -7 RR | -8 RR | -9 RR | 0 RR | 1 RR | 2 RR | 3 RR | | ర్ | | 5 RR | 6 RR | 7 RR | 8 RR | 9 RR | -20 RR | I RR | 2 RR | 3 RR | -24 RR | -25 RR |
| Resistances: CR | H12 | - 1112 | Plant# | | | · | • | | • | , | | -10 | -11 | -12 | -13 | | Plant # | | -15 | -16 | -17 | -18 | -19 | -2 | -21 | -22 | -23 | 7- | -2 |
| | 4 | T I | 8 | 1 | 7 | 3 | 4 | 5 | 9 | 7 | 90 | 6 | 10 | 11 | 12 | | Œ. | 1 | 2 | | 4 | w | 9 | 7 | 80 | 6 | 10 | 11 | 12 |
| MICROTITERPLATE # NL 05-403 | If coole which exertions: | rt seeds, waten positions: CR tieks tear (Ges/Dan | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| RPL | L cool | K II | ອ | 7. | Ħ | Þ | E | E | E | E | Ŀ | Ħ | E | RR | R | | ಕ | è | 푔 | RR | X | 訊 | R. | KR. | RR | RR | RR | RR | RR |
| ROTITE | Material: | Program : | Plant# | Green Forest | -26 | -27 | -28 | -29 | -30 | -31 | -32 | -33 | -34 | E16.LE157 | -2 | | Plant# | Ε, | 4 | · . | 9- | 1- | œ | 6- | -10 | -11 | -12 | -13 | -14 |
| | 2 | þΰ | | | ļ | | | | | | | | | | | l | | 1 | | | | | | ļ | | | | | |

| | MICROTITE <u>Material:</u> | PER. | MICROTITERPLATE # NIL 05-404 | | Z. | Resistances: CR | S | DNA-isolation: | | TAB | CTAB/Promega/Kac | 1/Kac | DNA-check: ves / no | Dat ck: ve | # | 24±05=2005 | NL (|
|----------|-------------------------------|----------|------------------------------|----------|-----|-----------------|-----|----------------|---|-----|------------------|--------|---------------------|---------------|-----------|-------------------------|--------|
| Program: | | H | If seeds, which positions: | attions: | [A] | <u>A1 - H12</u> | | | | | | | | ζ.]. | | | 05-404 |
| } | | 1 | | | | | | | | | | Ke | Kesults scored by: | | | <u>Date:</u> 00-00-0000 | |
| <u>a</u> | Plant# | <u> </u> | క్ | | B | Plant# | క | | | | C Plant# | # CR | | D | Plant# CR | | |
| ₹∣ | Augustus | | RR | | - | | | | | | 1 | л 7- | | 1 | | | |
| | -16 | | RR | | 2 | -27 | RR | | | , · | 7 | -5 IT | | 2 | -16 п | | |
| - 1 | -17 | | RR | | | -28 | RR | | | '. | ю | -6 п | | 60 | -17 ш | | |
| ļ | -18 | | RR | | 4 | -29 | RR | | | • | 4 | -7 п | | 4 | -18 | | |
| 1 | -19 | | RR | | ъ | -30 | Æ | | | | · vo | # 8- | | ın | п 61- | | |
| ! | -20 | | RR | | 9 | -31 | RR | | | | 9 | TI 6° | | 9 | -20 m | | |
| - 1 | -21 | | RR | | 7 | -32 | RR | | | | 7 | .10 п | | 7 | -21 ш | | |
| - 1 | -22 | | RR | | 8 | -33 | RR. | | | | | -11 ш | | 00 | -22 ш | | |
| - 1 | -23 | | RR | | 6 | -34 | RR | | | | 6 | .12 п | | 6 | -23 п | | |
| - 1 | -24 | | RR | | 10 | Green Towers | Ħ | | | - | 10 | -13 п | | 10 | .24 п | | |
| | -25 | | RR | | 11 | -2 | Ħ | | | 1 | 11 | -14 п | | = | -25 п | | |
| | -26 | | RR | | 12 | -3 | E | | | 1 | 12 | -15 пг | | 12 | -26 п | | |
| | | ŀ | | | | | Ì | | | | | | | | | | |
| Ы | Plant# | ٥ | CR | | i. | Plant# | రో | | | • | (Plant# | # CR | | H | Plant# CR | | |
| - 1 | -27 | | E | | - | 建设设置 | | | | | 1 | -14 RR | | 1 | | | |
| - 1 | -28 | | 6 | | 2 | .3 | RR | | | ۲ | 2 | -15 RR | | 7 | -26 RR | | |
| . | 29 | • | H | | 3 | 4 | RR | | | 64 | 3 | -16 RR | | m | -27 RR | | |
| | -30 | | н | | 4 | 5- | R. | | | 4 | 4 | -17 RR | | 4 | -28 RR | | |
| l | -31 | | н | | 2 | 9- | RR | | , | 41 | v. | -18 RR | | ıs. | -29 RR | | |
| | -32 | | ıı | 7 | 9 | | R. | | | • | 9 | -19 RR | | 9 | -30 RR | | |
| | -33 | | п | | 7 | 8- | Æ | | | • | J. | -20 RR | | 7 | -31 RR | | |
| | -34 | | E | | * | 6- | RR | | | ~ | 8 | -21 RR | | 8 | -32 RR | | |
| - 1 | -35 | | E | - | 6 | -10 | RR | | | | 6 | -22 RR | | 6 | -33 RR | | |
| | -36 | | × | | 10 | -11 | R. | | | 1 | 10 | -23 RR | - | 10 | -34 RR | | |
| 9 1 | Gladiator | | RR | | 11 | -12 | RR. | | | п | 1 | -24 RR | | 11 | -35 RR | | |
| | -2 | | RR | | 12 | -13 | RR | | | 1. | 12 | -25 RR | | 12 | -36 RR | | |

U.S. DEPARTMENT OF AGRICULTURE AGRICULTURAL MARKETING SERVICE SCIENCE DIVISION

| OBJECT | LETTUCE <u>Lactuca sativ</u> | VARIETY * | | |
|---|--|----------------------------------|---------------------|------------------------|
| NAME OF APPLICANT (S) | | | FOR OFF | ICIAL USE ONLY |
| Seminis Vegetable Seeds, I | Enc. Enza Zaden Be | eheer B.V. | VPONUMBER O | O O O / O |
| Go | P Code) | - | | 100249 |
| 37437 State Hwy 16 Woodladd, CA 95695 USA | | [· | ARIETY HAME | |
| Woodland CA 95695 USA | | | *Outback | |
| | | | XPERIMENTAL D | |
| Place numbers in the boxes for the characters which best desc spaced plants. Royal Horticultural Society or any recognized | ribe this variety. Measured di color standard may be used | ata should be the me | an of an appropriat | e-number (at least 10) |
| The location of the test area is: Narromine, NS | 17.7 | System Used: | | |
| Australia, & Huron, Califia | , | 773cm 0300: | | |
| 1. PLANT TYPE: (See list of suggested check varieties pa | age 4.j | | | |
| 01=Curting/Leaf 05=Grea 02=Butterhead 06=Vang 03=Bibb 07=Impe | t Lakes Group 09- guard Group 10- erial Group 10- | -Stern -Latin -OTHER- | | |
| 2. SEED: COLOR LIGHT | em (Ithaca) Group | | | |
| - Control Gray | The DORMANCY That Required F | HEAT DORM | ANCY | |
| 3=Brown (Amber) 2=Lio | ght Not Required | 2 1=Susceptible 2=Not Suscept | *t. 1 | |
| | | | ible | |
| 2 SHAPE OF COTYLEDONS: 1*Broad | ide a color photograph or ph in under optimal conditions. | otocopy of the fourt | 1 leaf from 20 day | old seedling |
| | 2=Intermediate | 3=Spatulate | | |
| 1 2 | \bigcup_{3} | | | |
| 2 0 LENGTH/WIDTH INDEX OF FOURTH LEAF: | - · · · · · · · · · · · · · · · · · · · | | 6 | |
| 1 APICAL MARGIN:) 1-Entire | 4=Moderately Den | itata 7 · | | |
| BASAL MARGIN: 2-Creanate/Gnawe | d 5=Coarsely Dentated | | | |
| 3=Finely Dentate | 6=Incised | - 0-O1 | HER (specify) | |
| 1 UNDULATION: 1=Flat | 2-Slight | 3 - Me | dium | 4=Marked |
| GREEN COLOR: 1=Yellow Green | 3=Medium Green | | | |
| 3 2-Light Green | 4=Dark Green | | e Green er Green | 7=Gray Green |
| ANTHOCYANIN: | | 0-3110 | -, Oreal | |
| DISTRIBUTION: 1=Absent 2=Margin Only | 3~Spotted 4~Throughout | 5 - 0Ti | HER (specify) | |
| CONCENTRATION: 1-Light | 2=Moderate | - 3=Inte | nse | |

2=Present

2=Slight

2=Apical Margin

3=Markedly

3*Lateral Margins

FORM LS-470-1 Formerly Form CR-470-1 which is obsolete (9-86)

1=Absent

1=Uncupped

1=None

ROLLING:

CUPPING:

REFLEXING:

| 4. MA | TURE | LEAVES (observe harvest-me | sture outer leaves): | | |
|----------|----------|--|--|--|---|
| | | | mature leaves which accurately shows color | and margin characteristics. 0 0 0 | 00249 |
| | 1 | INCISION DEPTH: Ideepest penetration o | 1=Absent/Shallow (Dark Green Boston f the margin) | n) 2=Moderate (Vanguard) | 3=Deep (Great Lakes 659 |
| | 2 | INDENTATION: (finest divisions of the mar | | 3-Defply Dentate (Great Lakes 659 4-Crenate (Vanguard) | 5-OTHER (specify) |
| | 1 | UNDULATION OF TI APICAL MARGIN: | 1=Absent/Slight (Dark Green Boston) | 2=Moderate (Vanguard) | 3-Strong (Great Lakes 65) |
| | 4 | GREEN COLOR: | 1=Very Light Green (Bibb) 2=Light Green (Minetto) | 3-Medium Green (Great Lakes) 4-Dark Green (Vanguard) | 5=Very Dark Green 6-OTHER |
| | | ANTHOCYANIN (grown a | t or below 10 C): | | |
| | | DISTRIBUTION: | 1=Absent 2=Margin Only (Big Boston) | 3=Spotted (Calif. Cream Butter) 4=Throughout (Prize Head) | 5-OTHER (*Pecify) |
| | | CONCENTRATION: | 1=Light (Iceberg) | 2=Moderate (Prize Head) | 3=Intense (Ruby) |
| | 3 | SIZE: | 1=Small | 2=Medium | 3=Large |
| | | GLOSSINESS: | 1=Dult (Vanguard) | 2=Moderate (Salinas) | 3=Glossy (Great Lakes) |
| | | BLISTERING: | 1=Absent/Slight (Salinas) | 2⇒Moderate (Vanguard) | 3≈Strong (Prize Head) |
| | | LEAF THICKNESS: | 1=Thin | 2=Intermediate | 3=Thick |
| | | TRICHOMES: | 1=Absent (smooth) | 2*Present (spiny) | |
| 5. PLAN | iT (at | market stage. Choose a comp | arison variety appropriate for this type.): | | |
| F- | ا | SPREAD OF FRAME LEAVE cm. This Variety | 1 1 1 | | |
| 12 | L_41. | W | 5 2 cm Green Towe | CS (specify comparison variety | y) |
| 3 | _ امــــ | cm This Variety | 2 9 cm Green Towe | TS (specify comparison variety | |
| JEM 4 | | HEAD SHAPE: | 1=Flattened 2=Slightly Flattened | 3=Spherical | 5=Non-Heading 6=OTHERROMaine |
| | 3 | HEAD SIZE CLASS: | 1=Small | 2=Medium | 3≖Large |
| 2 | 4 | HEAD COUNT PER CARTO | ٧ | | |
| 6 6 | I | HEAD WEIGHT: This Variety | 590 Green Towers | S | , |
| [| <u> </u> | HEAD FIRMNESS: | 1=Loose 2=Moderate | 3=Firm 4=Very Firm | |
| 6. BUTT | (botto | m of market-trimmed head); | | | |
| | 3 5 | SHAPE: | 1=Slightly Concave | 2=Flat | - 3=Rounded |
| 7 5085 (| <u> </u> | AIDRIB: | 1-Flattened (Salinas) | 2=Moderately Raised | 3=Prominently Raised (Great Lakes 659) |
| r | | f morket-trimmed head): | | | |
| | 3 _ " | nm Diameter at base of head | | | |
| 161 | <u> </u> | atio of head diameter/core dia ore height from base of head t | | v. | |
| 4 | ^ 1 | m This Variety | 4 7 mm Green Towe | CG (energify on the second | |
| BOLTIN | G (Ci | ve First Water Date | NOTE: First Water Date is to | te date seed first receives edequate main | ure |
| 5 | | | to perminate. This can and of | ten does equal the planting data. | · . |
| | <u> </u> | | -Very Slow | (specify comparison variety) | Very Rapid |
| | ਨਾ | A | 1-C1 | #Rapid | |
| | 5 cm | ight of mature seed stalk: This Variety | 8 4 Green Towers | (specify comparison variety) | |
| ORM LS-4 | 70-1 | | BOLTING cont'd, on next page | | |

| 1 BOUTER LEAVES: 1-5 | Straight | 2=Curved | | |
|--|---|---|--|--------|
| MARGIN: 1×0 | Sotire | 24Dentate | | |
| _2 color: ≽i | Sight Green | 2=Medium Green | 3-Dark Green | |
| BOLTER HABIT: | Absent | | men dengan kepada pada di ian pada mengabah bersada di Menjada di menjada di Kalendaran di Aleman di Alema Menjada di Aleman d | |
| . [1] LATERAL SHOOTS: | Noseni | 2=Present 2=Present | | |
| 1 BASAL SIDE SHOOTS: 3-4 | | 2-Present | | |
| MATURITY (carliness of harvest-mature head for | armattonj: | | | |
| NOTE: Camplete this section for at least on SEASON Applic, 1/ #of days C | | · CHECK V | | |
| Spring 5 2 | | | V YTSIRA | |
| Summer Summer | Gr | een Towers | | |
| | | | | |
| Spring o | 0 3 | een Towers | | |
| Sive planting date(s), and location(s): | | | | |
| Spring Narromine, NSW, | Australia"Tran | splanted" 15 S | ept. '99 | |
| Summer | | | | |
| SPRING Huron, Californ | ia - Sown 10 Janu | ary 2000 | | |
| | | | | |
| Winter 1/ First water date to hervesti | Z/Fill in check variety name on t | ns appropriate line. | | |
| U First water date to hervess. ADAPTATION: | | | | |
| LY FIRST WASTER DATE TO DEFVEST. ADAPTATION: PRIMARY REGIONS OF ADAPTIC | ON (tested and proven adapted): | (O-Not tested : | -Not Adapted 2-Adapted |)) |
| U First water date to hervess. ADAPTATION: PRIMARY REGIONS OF ADAPTIC 2 Southwest (Calif., Ark. desect) | ON (tested and proven adapted): 2 West Coast 2 | (O-Not tested | -Not Adapted 2-Adapted | 3) |
| D First water date to hervess. ADAPTATION: PRIMARY REGIONS OF ADAPTIC Southwest (Calif., Arts. desect) Northcentral SEASON: | ON (tested and proven adapted): 2 West Coast 2 Southeast | (O-Not tested 1 Northeast OTHER | | 3) |
| J First water date to hervess. ADAPTATION: PRIMARY REGIONS OF ADAPTIC 2 Southwest (Calif., Ark., decert) Northcentral SEASON: Spring (area_West_Coa | DN (tested and proven adapted): 2 West Coast Southeast St 1 | (G-Not tested 1 Northeast OTHER Fall (area NOTT | heast , |)) |
| J First water date to hervess. ADAPTATION: PRIMARY REGIONS OF ADAPTIC Southwest (Calif., Ark., desect) Northcentral SEASON: Spring larea_West_Coal Summer larea_West_Coal | ON (tested and proven adapted): 2 West Coast Southeast St , ast , | (G-Not tested : Northeast OTHERNort Fall (areaNort Winter (areaSouthw | heast 1 est 1 | |
| J First water date to hervett. ADAPTATION: PRIMARY REGIONS OF ADAPTIC 2 Southwest (Calif., Aris. desert) Northcentral SEASON: Spring larea West Coa Summer larea West Coa GREENHOUSE: 0-No | ON (tested and proven adapted): 2 West Coast Southeast St ast) (tested | (G-Not tested : Northeast OTHER Fall (area Nort Winter (area Southw | heast , est , | 33 |
| J First water date to hervess. ADAPTATION: PRIMARY REGIONS OF ADAPTIC Southwess (Calif., Arts. desert) Northcentral: SEASON: Spring (area West Coal Summer (area West Coal O GREENHOUSE: 0-No | ON (tested and proven adapted): 2 West Coast Southeast St ast) (tested | (G-Not tested : Northeast OTHERNort Fall (areaNort Winter (areaSouthw | heast 1 est 1 |)) |
| J First water date to hervess. ADAPTATION: PRIMARY REGIONS OF ADAPTIC 2 Southwest (Calif., Ark. desect) Northcentral: SEASON: Soring larea West Coa Summer larea West Co GREENHOUSE: 0-No | ON (tested and proven adapted): 2 West Coast Southeast St ast) (tested | (G-Not tested : Northeast OTHER Fall (area Nort Winter (area Southw | heast , est , | |
| J First water date to hervess. ADAPTATION: PRIMARY REGIONS OF ADAPTIC Southwess (Calif., Arts. desert) Northcentral: SEASON: Spring (area West Coal Summer (area West Coal O GREENHOUSE: 0-No | ON (tested and proven adapted): 2 West Coast Southeast St ast) (tested | (G-Not tested : Northeast OTHER Fall (area Nort Winter (area Southw | heast , est , | |
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11. DISEASES AND STRESS REACTIONS (0=Not tested; 1=Susceptible; 2=Intermediate; 3=Resistant; 4=Highly resistant; 5=Tolerant): VIRUS FUNGAL/BACTERIAL 1 Big Vein Corky Root Rot (Pythium Root Rot) Lattuca Mosaic Downy Mildew (Races Cucumber Mosaic Powdery Mildew Broad Bean Wilt Sclerotinia Rot Turnip Mossic Bacterial Soft Rot (Pseudomonas spp. & others) Beet Western Yellows Botrytis (Gray Mold) Lett. Infectious Yellows OTHER Corky Root (Rhizomonas Suberifaciens) Other Virus INSECTS PHYSIOLOGICAL/STRESS Cabbage Loopers Tipbum Salt Root Aphids A Brown Rib (Rib Discoloration, Rib Blight) Green Peach Aphid Drought OTHER Other Insect Cold POST HARVEST O Plak Rib Internal Rib Necrosis (Blackheart, Gray Rib, Gray Streak) n Russet Spotting Brown Stain O Rusty Brown Discoloration 12. BIOCHEMICAL OR ELECTROPHORETIC MARKERS: See Attached 13. COMMENTS: SUGGESTED CHECK VARIETIES TYPE

CUTTING/LEAF

BUTTERHEAD

31 8188

COS, OR ROMAINE

5) GREAT LAKES GROUP

6) 7) VANGUARD GROUP

IMPERIAL GROUP 8) EASTERN GROUP

91 STEM

LATIN

CHECK VARIETY

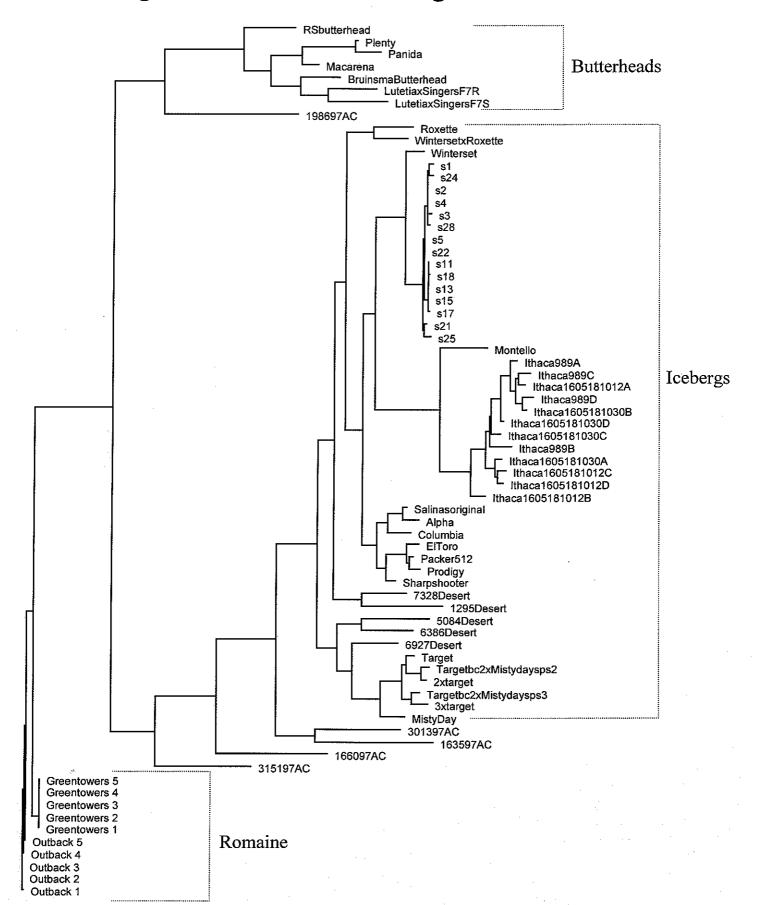
SALAD BOWL DARK GREEN BOSTON 8818 PARRIS ISLAND GREAT LAKES 659-700 VANGUARD VIVA ITHACA CELTUCE MATCHLESS

Item 12. Biochemical Data

Figure 1: A rooted dendrogram using PAUP software indicating the genetic distance among varieties.

The dendrogram indicates the genetic distance between and within three groups of lettuce – Butterhead, Iceberg and Romaine.

The dendrogram highlights that Outback (OB 1-5) differs from Green Towers (GT 1-5) using microsatellite technology.



The following statements are made in accordance with the Privacy Act of AGRICULTURAL MARKETING SERVICE 1974 (5 U.S.C. 552a) and the Paperwork Reduction Act (PRA) of 1995. **EXHIBIT E** Application is required in order to determine if a plant variety protection certificate is to be issued (7 U.S.C. 2421). Information is held confidential STATEMENT OF THE BASIS OF OWNERSHIP until certificate is issued (7 U.S.C. 2426). 1. NAME OF APPLICANT(S) 2. TEMPORARY DESIGNATION 3. VARIETY NAME OR EXPERIMENTAL NUMBER Yates ∀egetable Seeds Enza Zaden Beheer B.V. Outback 4. ADDRESS (Street and No., or R.F.D. No., City, State, and ZIP, and Country) 5. TELEPHONE (include area code) 6. FAX (include area code) Gerrit Sassen 13/19-Chifley Street 61297251066-00.31,228,351318 Enza Zaden Beheer B.V. 7. PVPO NUMBER Smithfield 2164-AUS Enkhuizen The Netherlands 8. Does the applicant own all rights to the variety? Mark an "X" in appropriate block. If no, please explain. pAD 10/27/05 Is the applicant (individual or company) a U.S. national or U.S. based company? YES NO -Australia If no, give name of country Netherlands 10. Is the applicant the original owner? If no, please answer one of the following: a. If original rights to variety were owned by individual(s), is (are) the original owner(s) a U.S. national(s)? If no, give name of country ┐YES b. If original rights to variety were owned by a company(ies), is(are) the original owner(s) a U.S. based company? NO If no, give name of country 11. Additional explanation on ownership (if needed, use reverse for extra space): PLEASE NOTE: Plant variety protection can be afforded only to owners (not licensees) who meet one of the following criteria: 1. If the rights to the variety are owned by the original breeder, that person must be a U.S. national, national of a UPOV member country, or national of a country which affords similar protection to nationals of the U.S. for the same genus and species. 2. If the rights to the variety are owned by the company which employed the original breeder(s), the company must be U.S. based, owned by nationals of a UPOV member country, or owned by nationals of a country which affords similar protection to nationals of the U.S. for the same genus and species. 3. If the applicant is an owner who is not the original owner, both the original owner and the applicant must meet one of the above criteria. The original breeder/owner may be the individual or company who directed final breeding. See Section 41(a)(2) of the Plant Variety Protection Act for definition. According to the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is OS81-00SS. The time required to compete this information collection is estimated to average 10 minutes per response, including the time for reviewing instructions, searching existing data sources, gathering and mainteining the data needed, and completing and reviewing the collection of information. The U.S. Department of Agriculture (USDA) prohibits discrimination in its programs on the basis of race, color, national origin, sex, religion, age, disability, political beliefs, and marital or familial status. (Not all prohibited bases apply to all programs). Persons with disabilities who require alternative means for communication of program information (braille, large print, audiotape, etc.) should contact

To file a compleint, write the Secretary of Agriculture, U.S. Department of Agriculture, Washington, D.C. 20250, or call 1-800-245-6340 (voice) or (202) 720-1127 (TDD). USDA is an equal

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employment opportunity employer.

USDA's TARGET Center at 202-720-2600 (voice and TDD).

Electronic version designed using WordPerfect InForms by USDA-AMS-IMB.

U.S. DEPARTMENT OF AGRICULTURE

| Form ST- (06/03) | RECORDATI for Plant Variet Offic | ty Protection | U.S. DEPARTMENT OF AGRICULTURE PLANT VARIETY PROTECTION OFFICE |
|---|--|--|---|
| To the Commissioner of Plant Va | riety Protection: Please | record the attached | d original documents or copy thereof. |
| 1. Current Owner(s) of Record | - | 2. Type of Recor | rdation: |
| Name Yates Vegetable Address 13-19 Chifley Smithfield 21 Australia Phone FAX E-mail | Street 164 | Merger Change of N Revocation Change of A Change of R Change of A Change of V Election of Other (speci | rerest, License, Grant, Conveyance Name of Owner(s) of Assignment, Security Interest, License, Grant, or Conveyance Address of Owner(s) Representative (and address) Address of Representative Variety Name (Denomination) "Certified Seed Only" Option ify) |
| 3. New Owner(s) | | 4. New Represen | itative |
| Name Enza Zaden Be | heer B.V. | Name | |
| Address P.O. Box 7 160 | 00 AA | Address | |
| Haling 1e, 1602 | 2 DB | | |
| Enkhuizen, The No | etherlands | | - Commence |
| Phone 00 31 228 35 | 01.00 | Phone | |
| FAX 00 31 228 31 | 5960 | FAX | |
| B-mail info@enzazado | en.nl | E-mail | |
| 7. List PVP Number(s), Crop Ki [Please note that listed applications and center of the company o | ertificates must be active. Rei | cordations cannot be per | |

8. Total number of applications/certificates involved: _____ Total Fee (97.175) ________

Fees must be paid in U.S. funds. Make checks payable to "Treasurer of the United States". We cannot accept payment by electronic fund transfer or credit card.

9. Statement and Signature

To the best of my knowledge and belief, the fargoing influence and any attached copy is a true copy of the original document.

Map lambalk
Name of Person Signing

1600 AA Ehkhuizen

28-July-2005 Date

Mail documents to: Plant Variety Protection Office

NAL Building, Room 400 10301 Baltimore Blvd. Beltsville, MD 20705-2351